

10/081,455

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(FILE 'HOME' ENTERED AT 07:27:21 ON 02 JUN 2005)

INDEX 'ADISCTI, ADISINSIGHT, ADISNEWS, AGRICOLA, ANABSTR, ANTE, AQUALINE,
AQUASCI, BIOBUSINESS, BIOCOMMERCE, BIOENG, BIOSIS, BIOTECHABS, BIOTECHDS,
BIOTECHNO, CABA, CANCERLIT, CAPLUS, CEABA-VTB, CEN, CIN, CONFSCI, CROPB,
CROPU, DDFB, DDFU, DGENE, DISSABS, ...' ENTERED AT 07:27:32 ON 02 JUN 2005

SEA SIALYLTRANSFERASE

9 FILE ADISCTI
1 FILE ADISINSIGHT
37 FILE AGRICOLA
20 FILE ANABSTR
8 FILE AQUASCI
11 FILE BIOBUSINESS
13 FILE BIOCOMMERCE
49 FILE BIOENG
1893 FILE BIOSIS
198 FILE BIOTECHABS
198 FILE BIOTECHDS
855 FILE BIOTECHNO
104 FILE CABA
693 FILE CANCERLIT
2312 FILE CAPLUS
40 FILE CEABA-VTB
2 FILE CEN
4 FILE CIN
58 FILE CONFSCI
24 FILE DDFB
46 FILE DDFU
809 FILE DGENE
98 FILE DISSABS
24 FILE DRUGB
53 FILE DRUGU
10 FILE EMBAL
1630 FILE EMBASE
663 FILE ESBIOBASE
33 FILE FEDRIP
5 FILE FROSTI
6 FILE FSTA
1491 FILE GENBANK
164 FILE IFIPAT
285 FILE JICST-EPLUS
424 FILE LIFESCI
1964 FILE MEDLINE
2 FILE NIOSHTIC
5 FILE NTIS
1 FILE OCEAN
651 FILE PASCAL
1 FILE PHIN
10 FILE PROMT
2 FILE PROUSDDR
1746 FILE SCISEARCH
598 FILE TOXCENTER
756 FILE USPATFULL
27 FILE USPAT2
110 FILE WPIDS
1 FILE WPIFV
110 FILE WPINDEX

QUE SIALYLTRANSFERASE

L1

FILE 'CAPLUS, MEDLINE, BIOSIS, SCISEARCH, EMBASE, BIOTECHNO, DGENE,
USPATFULL, CANCERLIT, ESBIOBASE, PASCAL, TOXCENTER, LIFESCI' ENTERED AT
07:28:35 ON 02 JUN 2005

L2 97 S L1 AND (PHOTOBACTERIUM DAMSELA)
L3 53 S L2 AND (ALPHA 2,6)
L4 32 DUP REM L3 (21 DUPLICATES REMOVED)
L5 367 S L1 AND (NEISSERIA MENINGITIDIS)
L6 173 S L5 AND (ALPHA 2,3)
L7 97 DUP REM L6 (76 DUPLICATES REMOVED)
L8 193 S L1 AND (CAMPYLOBACTER JEJUNI)
L9 69 S L8 AND (ALPHA 2,3)
L10 56 DUP REM L9 (13 DUPLICATES REMOVED)

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L7 ANSWER 87 OF 97 CAPLUS COPYRIGHT 2005 ACS on STN DUPLICATE 4
ACCESSION NUMBER: 1999:83325 CAPLUS
DOCUMENT NUMBER: 130:264522
TITLE: Role of lipopolysaccharide sialylation in serum resistance of serogroup B and C meningococcal disease isolates
AUTHOR(S): Vogel, Ulrich; Claus, Heike; Heinze, Gabriele; Frosch, Matthias
CORPORATE SOURCE: Institut fur Hygiene und Mikrobiologie, Universitat Wurzburg, Wurzburg, 97080, Germany
SOURCE: Infection and Immunity (1999), 67(2), 954-957
CODEN: INFIBR; ISSN: 0019-9567
PUBLISHER: American Society for Microbiology
DOCUMENT TYPE: Journal
LANGUAGE: English
AB ***.alpha.-2,3-Sialyltransferase***
mutants of 3 genetically and phenotypically diverse ***Neisseria meningitidis*** strains were compared with regard to resistance to human serum and systemic spread in the infant rat. Lipopolysaccharide sialylation was of minor importance for the resistance of serogroup B and C meningococcal disease isolates to complement attack.
REFERENCE COUNT: 31 THERE ARE 31 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 88 OF 97 USPATFULL on STN
ACCESSION NUMBER: 1998:157502 USPATFULL
TITLE: Isolated nucleic acid molecules which hybridize to polysialyl transferases
INVENTOR(S): Gerardy-Schahn, Rita, Hiddenhausen, Germany, Federal Republic of
Fukuda, Minoru, San Diego, CA, United States
Nakayama, Jun, Matsumoto, Japan
Eckhardt, Matthias, Hanover, Germany, Federal Republic of
PATENT ASSIGNEE(S): Boehringer Mannheim GmbH, Mannheim, Germany, Federal Republic of (non-U.S. corporation)
La Jolla Cancer Research Foun., La Jolla, CA, United States (U.S. corporation)

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 5849904		19981215
APPLICATION INFO.:	US 1995-576775		19951221 (8)
RELATED APPLN. INFO.:	Continuation-in-part of Ser. No. US 1995-503133, filed on 17 Jul 1995		

	NUMBER	DATE
PRIORITY INFORMATION:	DE 1995-116387	19951018
DOCUMENT TYPE:	Utility	
FILE SEGMENT:	Granted	
PRIMARY EXAMINER:	LeGuyader, John L.	
ASSISTANT EXAMINER:	Wang, Andrew	
LEGAL REPRESENTATIVE:	Felfe & Lynch	
NUMBER OF CLAIMS:	8	
EXEMPLARY CLAIM:	1	
NUMBER OF DRAWINGS:	23 Drawing Figure(s); 7 Drawing Page(s)	
LINE COUNT:	1969	

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

AB Isolated nucleic acid molecules encoding polysialyl transferases, and the polysialyl transferases themselves are disclosed. SEQ ID NOS: 1, 2, 7 and 8 present examples of these. The nucleic acid molecules and the

proteins can be used diagnostically or therapeutically. Additionally, antisense oligonucleotides and antibodies are described, which can also be used diagnostically or therapeutically.

L7 ANSWER 89 OF 97 CAPLUS COPYRIGHT 2005 ACS on STN DUPLICATE 5
ACCESSION NUMBER: 1998:784305 CAPLUS
DOCUMENT NUMBER: 130:152327
TITLE: The (α 2 \rightarrow 8)-linked polysialic acid capsule and lipooligosaccharide structure both contribute to the ability of serogroup B *Neisseria meningitidis* to resist the bactericidal activity of normal human serum
AUTHOR(S): Kahler, C. M.; Martin, L. E.; Shih, G. C.; Rahman, M. M.; Carlson, R. W.; Stephens, D. S.
CORPORATE SOURCE: Department of Medicine, Emory University School of Medicine, Atlanta, GA, 30033, USA
SOURCE: Infection and Immunity (1998), 66(12), 5939-5947
CODEN: INFIBR; ISSN: 0019-9567
PUBLISHER: American Society for Microbiology
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The mol. basis for the resistance of serogroup B *Neisseria meningitidis* to the bactericidal activity of normal human sera (NHS) was examined with a NHS-resistant, invasive serogroup B meningococcal isolate and genetically and structurally defined capsule-, lipooligosaccharide (LOS)-, and sialylation-altered mutants of the wild-type strain. Expression of the (α 2 \rightarrow 8)-linked polysialic acid serogroup B capsule was essential for meningococcal resistance to NHS. The very NHS-sensitive phenotype of acapsular mutants (99.9 to 100% killed in 10, 25, and 50% NHS) was not rescued by complete LOS sialylation or changes in LOS structure. However, expression of the capsule was necessary but not sufficient for a fully NHS-resistant phenotype. In an encapsulated background, loss of LOS sialylation by interrupting the alpha.2,3 sialyltransferase gene, 1st, increased sensitivity to 50% NHS. In contrast, replacement of the lacto-N-neotetraose α -chain (Gal β 1-4GlcNAc β 1-3Gal β 1-4Glc) with glucose extensions (GlcN) in a galE mutant resulted in a strain resistant to killing by 50% NHS at all time points. Encapsulated meningococci expressing a Hep2(GlcNAc) \rightarrow KDO2 \rightarrow lipid A LOS without an α -chain demonstrated enhanced sensitivity to 50% NHS (98% killed at 30 min) mediated through the antibody-dependent classical complement pathway. Encapsulated LOS mutants expressing truncated Hep2 \rightarrow KDO2 \rightarrow lipid A and KDO2 \rightarrow lipid A structures were also sensitive to 50% NHS (98 to 100% killed at 30 min) but, unlike the wild-type strain and mutants with larger oligosaccharide structures, they were killed by hypogammaglobulinemic sera. These data indicate that encapsulation is essential but that the LOS structure contributes to the ability of serogroup B *N. meningitidis* to resist the bactericidal activity of NHS.
REFERENCE COUNT: 70 THERE ARE 70 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 90 OF 97 CAPLUS COPYRIGHT 2005 ACS on STN DUPLICATE 6
ACCESSION NUMBER: 1998:510061 CAPLUS
DOCUMENT NUMBER: 129:255694
TITLE: The synthesis of sialylated oligosaccharides using a CMP-Neu5Ac synthetase/sialyltransferase fusion
AUTHOR(S): Gilbert, Michel; Bayer, Robert; Cunningham, Anna-Marie; DeFrees, Shawn; Gao, Yinghong; Watson, David C.; Young, N. Martin; Wakarchuk, Warren W.
CORPORATE SOURCE: Institute for Biological Sciences, National Research Council of Canada, Ottawa, ON, K1A 0R6, Can.

SOURCE: Nature Biotechnology (1998), 16(8), 769-772
CODEN: NABIF9; ISSN: 1087-0156

PUBLISHER: Nature America

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Large-scale enzymic synthesis of oligosaccharides, which contain terminal N-acetyl-neuraminic acid residues requires large amts. of the **sialyltransferase** and the corresponding sugar-nucleotide synthetase, which is required for the synthesis of the sugar-nucleotide donor, CMP-Neu5Ac. Using genes cloned from **Neisseria meningitidis**, we constructed a fusion protein that has both CMP-Neu5Ac synthetase and **alpha**-2,3-sialyltransferase activities. The fusion protein was produced in high yields (over 1200 U/L, measured using an **alpha**-2,3-sialyltransferase assay) in *Escherichia coli* and functionally pure enzyme could be obtained using a simple protocol. In small-scale enzymic syntheses, the fusion protein could sialylate various oligosaccharide acceptors (branched and linear) with N-acetyl-neuraminic acid as well as N-glycolyl- and N-propionyl-neuraminic acid in high conversion yield. The fusion protein was also used to produce **alpha**-2,3-sialyllactose at the 100 g scale using a sugar nucleotide cycle reaction, starting from lactose, sialic acid, phosphoenolpyruvate, and catalytic amts. of ATP and CMP.

REFERENCE COUNT: 16 THERE ARE 16 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 91 OF 97 LIFESCI COPYRIGHT 2005 CSA on STN
ACCESSION NUMBER: 1998:102400 LIFESCI
TITLE: Sweet success with tethered enzyme catalysis
AUTHOR: Warner, T.G.
CORPORATE SOURCE: Genentech, 1 DNA Way, South San Francisco, CA 94080, USA
SOURCE: Nat. Biotechnol., (19980800) vol. 16, no. 8, pp. 720-721.
ISSN: 1087-0156.

DOCUMENT TYPE: Journal
TREATMENT CODE: General Review
FILE SEGMENT: W3
LANGUAGE: English

AB The synthesis of complex carbohydrates can often seem a daunting task. It requires not only a series of complex chemical syntheses, but also meticulous attention to the proper stereochemical orientation of the sugar molecules at each step of the process. Even a simple carbohydrate composed of four different sugar units has an estimated 34,560 possible isomeric variations. Creating a simple glycan molecule is thus a Herculean challenge for all but the most daring of synthetic chemists. In this issue, help may be on the way in the form of a novel enzymatic approach described by Gilbert and coworkers. They show that their bifunctional enzyme, a fusion of two microbial enzymes, can both simplify the production and purification of reagent enzymes and facilitate oligosaccharide synthesis.

L7 ANSWER 92 OF 97 CAPLUS COPYRIGHT 2005 ACS on STN DUPLICATE 7
ACCESSION NUMBER: 1998:433863 CAPLUS
DOCUMENT NUMBER: 129:174408
TITLE: Structure of an α -2,6-sialylated lipooligosaccharide from **Neisseria meningitidis** immunotype L1
AUTHOR(S): Wakarchuk, Warren W.; Gilbert, Michel; Martin, Adele; Wu, Yuyang; Brisson, Jean-Robert; Thibault, Pierre; Richards, James C.
CORPORATE SOURCE: Institute for Biological Sciences, National Research Council of Canada, Ottawa, ON, K1A 0R6, Can.
SOURCE: European Journal of Biochemistry (1998), 254(3), 626-633

CODEN: EJBCAI; ISSN: 0014-2956

PUBLISHER: Springer-Verlag
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The recent cloning of the lipooligosaccharide (LOS) α -2,3-sialyltransferase from *Neisseria meningitidis* immunotype L3 permitted us to examine other immunotypes for this structural gene. We identified the gene and measured the enzyme activity in the L1 immunotype strain which had previously been reported to lack sialic acid in its LOS because it contains a terminal α -linked galactose which was thought not to be an acceptor for the sialyltransferase. This finding prompted us to re-examine the structure of the LOS from the L1 immunotype, which revealed the presence of sialic acid on the terminal α -linked galactose. Oligosaccharides derived from the LOS were shown to be sialylated by composition and methylation anal., mass spectrometry and NMR. The detailed structural anal. showed the sialic acid to occur only at O6 of the terminal α -D-galactopyranose residue of the α -D-Gal-1,4- β -D-Gal-1,4- β -D-gluc trisaccharide (Pk epitope) chain of the LOS, in the α -D configuration. These data are the first report of an α -2,6-linked sialic acid in a bacterial LOS or lipopolysaccharide, and also the first report of a sialylated Pk epitope.

REFERENCE COUNT: 34 THERE ARE 34 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 93 OF 97 CAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1998:15851 CAPLUS
DOCUMENT NUMBER: 128:71644
TITLE: Cloning and substrate specificity of *Neisseria* recombinant α -2,3-sialyltransferases
INVENTOR(S): Gilbert, Michel; Wakarchuk, Warren W.; Young, N. Martin; Jennings, Michael P.
PATENT ASSIGNEE(S): National Research Council of Canada, Can.
SOURCE: PCT Int. Appl., 50 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9747749	A1	19971218	WO 1997-CA390	19970610
W: CA, FI, JP, NO				
RW: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
US 6096529	A	20000801	US 1997-872485	19970607
CA 2256645	AA	19971218	CA 1997-2256645	19970610
EP 906432	A1	19990407	EP 1997-923695	19970610
EP 906432	B1	20040609		
R: CH, DE, DK, ES, FR, GB, IT, LI, SE, IE, FI				
JP 2001503961	T2	20010327	JP 1997-526320	19970610
US 6210933	B1	20010403	US 1999-387942	19990901
PRIORITY APPLN. INFO.:			US 1996-19520P	P 19960610
			US 1997-872485	A 19970606
			WO 1997-CA390	W 19970610

AB The structure and specificity of the recombinant α -2,3-sialyltransferases from *Neisseria* spp. are disclosed. The genes encoding the α -2,3-sialyltransferases involved in lipooligosaccharide biosynthesis from *N. meningitidis* and *N. gonorrhoeae* were cloned and expressed in *Escherichia coli*. A high sensitivity enzyme assay using a synthetic fluorescent glycosyltransferase acceptor and capillary electrophoresis was

used to screen a genomic library of *N. meningitidis* MC58 L3 in a "divide and conquer" strategy. The gene, denoted 1st, was found on a 2.0-kb fragment of DNA, and its sequence was determined and then used to design probes to amplify and subsequently clone the corresponding 1st genes from *N. meningitidis* 406Y L3, *N. meningitidis* M982B L7, and *N. gonorrhoeae* F62. Functional sialyltransferase was produced from the genes derived from both L3 *N. meningitidis* strains and *N. gonorrhoeae* F62. However, the *N. meningitidis* M982B L7 gene contained a frameshift mutation that renders it inactive. The expression of the 1st gene was easily detected using the enzyme assay, and the protein expression could be detected when an immunodetection tag was added to the C-terminal end of the protein. Using the synthetic acceptor N-acetyllactosamineaminophenyl-6-(5-(fluorescein-carboxamido)-hexanoic acid amide), the α -2,3-specificity of the enzyme was confirmed by NMR examination of the reaction product. The enzyme could also use synthetic acceptors with lactose or galactose as the saccharide portion. The use of the enzymes in the production of desired carbohydrate structures is also provided.

L7 ANSWER 94 OF 97 CAPLUS COPYRIGHT 2005 ACS on STN DUPLICATE 8
ACCESSION NUMBER: 1997:375025 CAPLUS
DOCUMENT NUMBER: 127:105886
TITLE: Purification and characterization of the recombinant CMP-sialic acid synthetase from *Neisseria meningitidis*
AUTHOR(S): Gilbert, Michel; Watson, David C.; Wakarchuk, Warren W.
CORPORATE SOURCE: Inst. Biological Sci., National Res. Council Canada, Ottawa, ON, K1A 0R6, Can.
SOURCE: Biotechnology Letters (1997), 19(5), 417-420
CODEN: BILED3; ISSN: 0141-5492
PUBLISHER: Chapman and Hall
DOCUMENT TYPE: Journal
LANGUAGE: English
AB CMP-Sialic acid synthetase from *Neisseria meningitidis* 406Y was expressed in *Escherichia coli* K113 pLysS and produced at 360 U/L. The purified CMP-sialic acid synthetase used both N-acetyl-neuraminic acid ($K_m=0.34$ mM) and N-glycolyl-neuraminic acid ($K_m=2.6$ mM) as substrates. The recombinant synthetase could be used in a coupled reaction with an α -2,3-sialyltransferase to sialylate a lactose derivative in a one-reactor synthesis.
REFERENCE COUNT: 11 THERE ARE 11 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 95 OF 97 CAPLUS COPYRIGHT 2005 ACS on STN DUPLICATE 9
ACCESSION NUMBER: 1997:686481 CAPLUS
DOCUMENT NUMBER: 127:356419
TITLE: Characterization of a recombinant *Neisseria meningitidis* α -2,3-sialyltransferase and its acceptor specificity
AUTHOR(S): Gilbert, Michel; Cunningham, Anna-Maria; Watson, David C.; Martin, Adele; Richards, James C.; Wakarchuk, Warren W.
CORPORATE SOURCE: Institute for Biological Sciences, National Research Council of Canada, Ottawa, ON, K1A 0R6, Can.
SOURCE: European Journal of Biochemistry (1997), 249(1), 187-194
CODEN: EJBCAI; ISSN: 0014-2956
PUBLISHER: Springer
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The structure and specificity of the recombinant α -2,3-sialyltransferase from *Neisseria*

meningitidis are reported. This enzyme showed an unusual acceptor specificity in that it could use α -terminal and β -terminal Gal residues as acceptors. In addition ($\beta 1 \rightarrow 4$)-linked and ($\beta 1 \rightarrow 3$)-linked terminal Gal served as acceptors. These properties distinguish the bacterial enzyme from the more widely investigated mammalian equivalent. The protein was expressed as a membrane-associated protein in *Escherichia coli* at a level of 750 U/l (≈ 250 mg/l). The protein could be extracted with buffers containing 0.2% Triton X-100 and purified to homogeneity using immobilized-metal-affinity chromatog. Electrospray-ionization mass spectrometry of peptides obtained by cleavage with cyanogen bromide and trypsin confirmed over 95% of the deduced amino acid sequence. When used for enzymic synthesis in coupled reactions with recombinant CMP-Neu5Ac synthetase, the **.alpha.-2,3-sialyltransferase** could sialylate fluorescent derivs. of N-acetyllactosamine with N-acetylneuraminic acid, N-propionylneuraminic acid and N-glycoloylneuraminic acid.

REFERENCE COUNT: 29 THERE ARE 29 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 96 OF 97 CAPLUS COPYRIGHT 2005 ACS on STN DUPLICATE 10
ACCESSION NUMBER: 1997:687094 CAPLUS
DOCUMENT NUMBER: 128:47176
TITLE: Functional characterization of an isogenic meningococcal **.alpha.-2,3-sialyltransferase** mutant: the role of lipooligosaccharide sialylation for serum resistance in serogroup B meningococci
AUTHOR(S): Vogel, U.; Claus, Heike; Heinze, Gabriele; Frosch, Matthias
CORPORATE SOURCE: Institut fur Hygiene und Mikrobiologie, Universitat Wurzburg, Josef-Schneider-Strasse 2, Wurzburg, D-97080, Germany
SOURCE: Medical Microbiology and Immunology (1997), 186(2-3), 159-166
CODEN: MMIYAO; ISSN: 0300-8584
PUBLISHER: Springer
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The neisserial **.alpha.-2,3-sialyltransferase**, which is encoded by the 1st gene, terminally links sialic acid to the lacto-N-neotetraose residue of neisserial lipooligosaccharide (LOS). We used the recently published nucleotide sequence of the neisserial 1st gene to construct an isogenic serogroup B meningococcal 1st mutant by insertion of a kanamycin resistance gene. The resulting 1st mutant expressed the unsialylated lacto-N-neotetraose structure. Using bactericidal assays and an infant rat model of meningococcal infection, we were able to demonstrate that 1st mutation, in contrast to galE mutation, which results in a truncated LOS, or to siaD mutation, which results in loss of the capsule, neither had an effect on resistance to normal human serum, nor did it impair the ability of meningococci to spread systemically in the non-immune host. The 1st mutant was serum resistant despite of the fact that the central factor of complement activation, C3b, was deposited on the 1st mutant as efficiently as it was on the galE mutant. Thus, the terminal sialic acid residue linked to the wild-type LOS inhibited C3b deposition on the meningococcus. However, in contrast to the galE mutant, where C3b deposition is promoted by IgM binding, the 1st mutant's surface is not a target for IgM mols. Thus, the lacto-N-neotetraose residue of neisserial LOS alone, without the presence of terminal sialic acid, is sufficient to block IgM epitopes either on the LOS itself, or on other surface mols. Our data provide further insight into the complex interplay of capsular and LOS sialic acids in serogroup B meningococci with host effector mechanisms, and suggest that LOS sialylation in meningococci is of a less central

importance as it is in gonococci.

L7 ANSWER 97 OF 97 CAPLUS COPYRIGHT 2005 ACS on STN DUPLICATE 11

ACCESSION NUMBER: 1996:681790 CAPLUS

DOCUMENT NUMBER: 126:15318

TITLE: Cloning of the lipooligosaccharide α .
 α .-2,3-sialyltransferase from
the bacterial pathogens *Neisseria*
meningitidis and *Neisseria gonorrhoeae*

AUTHOR(S): Gilbert, Michel; Watson, David C.; Cunningham,
Anna-Maria; Jennings, Michael P.; Young, N. Martin;
Wakarchuk, Warren W.

CORPORATE SOURCE: Institute Biological Sciences, National Research
Council Canada, Ottawa, ON, K1A 0R6, Can.

SOURCE: Journal of Biological Chemistry (1996), 271(45),
28271-28276

CODEN: JBCHA3; ISSN: 0021-9258

PUBLISHER: American Society for Biochemistry and Molecular
Biology

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The genes encoding the α .-2,3-sialyltransferases involved in lipooligosaccharide biosynthesis from *Neisseria meningitidis* and *Neisseria gonorrhoeae* have been cloned and expressed in *Escherichia coli*. A high sensitivity enzyme assay using a synthetic fluorescent glycosyltransferase acceptor and capillary electrophoresis was used to screen a genomic library of *N. meningitidis* MC58 L3 in a "divide and conquer" strategy. The gene, denoted 1st, was found on a 2.0-kilobase fragment of DNA, and its sequence was determined and then used to design probes to amplify and subsequently clone the corresponding 1st genes from *N. meningitidis* 406Y L3, *N. meningitidis* M982B L7, and *N. gonorrhoeae* F62. Functional sialyltransferase was produced from the genes derived from both *N. meningitidis* strains and the *N. gonorrhoeae* F62. However, the *N. meningitidis* M982B L7 gene contained a frameshift mutation that renders it inactive. The expression of the 1st gene was easily detected using the enzyme assay, and the protein expression could be detected when an immunodetection tag was added to the COOH-terminal end of the protein. Using the synthetic acceptor N-acetyllactosamineaminophenyl-6-(5-(fluorescein-carboxamido)-hexanoic acid amide), the α .-2,3-specificity of the enzyme was confirmed by NMR examination of the reaction product. The enzyme could also use synthetic acceptors with lactose or galactose as the saccharide portion. This study is the first example of the cloning, expression, and examination of α .-2,3-sialyltransferase activity from a bacterial source.

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L10 ANSWER 46 OF 56 USPATFULL on STN
ACCESSION NUMBER: 2002:287597 USPATFULL
TITLE: Practical in vitro sialylation of recombinant glycoproteins
INVENTOR(S): Paulson, James C., Del Mar, CA, UNITED STATES
Bayer, Robert J., San Diego, CA, UNITED STATES
Sjoberg, Eric, San Diego, CA, UNITED STATES
PATENT ASSIGNEE(S): Neose Technologies, Inc., Horsham, PA (U.S. corporation)

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 2002160460	A1	20021031
APPLICATION INFO.:	US 2002-81456	A1	20020221 (10)
RELATED APPLN. INFO.:	Continuation of Ser. No. US 1998-7741, filed on 15 Jan 1998, GRANTED, Pat. No. US 6399336		

	NUMBER	DATE
PRIORITY INFORMATION:	US 1997-35710P	19970116 (60)
DOCUMENT TYPE:	Utility	
FILE SEGMENT:	APPLICATION	
LEGAL REPRESENTATIVE:	TOWNSEND AND TOWNSEND AND CREW, LLP, TWO EMBARCADERO CENTER, EIGHTH FLOOR, SAN FRANCISCO, CA, 94111-3834	
NUMBER OF CLAIMS:	58	
EXEMPLARY CLAIM:	1	
NUMBER OF DRAWINGS:	2 Drawing Page(s)	
LINE COUNT:	1142	

CAS INDEXING IS AVAILABLE FOR THIS PATENT.
AB This invention provides methods for practical in vitro sialylation of glycoproteins, including recombinantly produced glycoproteins. The methods are useful for large-scale modification of sialylation patterns.

L10 ANSWER 47 OF 56 USPATFULL on STN
ACCESSION NUMBER: 2002:258800 USPATFULL
TITLE: Practical in vitro sialylation of recombinant glycoproteins
INVENTOR(S): Paulson, James C., Del Mar, CA, UNITED STATES
Bayer, Robert J., San Diego, CA, UNITED STATES
Sjoberg, Eric, San Diego, CA, UNITED STATES
PATENT ASSIGNEE(S): Neose Technologies, Inc., Horsham, PA, UNITED STATES, 19044 (U.S. corporation)

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 2002142370	A1	20021003
APPLICATION INFO.:	US 2002-81455	A1	20020221 (10)
RELATED APPLN. INFO.:	Continuation of Ser. No. US 1998-7741, filed on 15 Jan 1998, GRANTED, Pat. No. US 6399336		

	NUMBER	DATE
PRIORITY INFORMATION:	US 1997-35710P	19970116 (60)
DOCUMENT TYPE:	Utility	
FILE SEGMENT:	APPLICATION	
LEGAL REPRESENTATIVE:	TOWNSEND AND TOWNSEND AND CREW, LLP, TWO EMBARCADERO CENTER, EIGHTH FLOOR, SAN FRANCISCO, CA, 94111-3834	
NUMBER OF CLAIMS:	58	
EXEMPLARY CLAIM:	1	
NUMBER OF DRAWINGS:	2 Drawing Page(s)	
LINE COUNT:	1135	

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

AB This invention provides methods for practical in vitro sialylation of glycoproteins, including recombinantly produced glycoproteins. The methods are useful for large-scale modification of sialylation patterns.

L10 ANSWER 48 OF 56 USPATFULL on STN
ACCESSION NUMBER: 2002:221376 USPATFULL
TITLE: Practical in vitro sialylation of recombinant glycoproteins
INVENTOR(S): Paulson, James C., Del Mar, CA, UNITED STATES
Bayer, Robert J., San Diego, CA, UNITED STATES
Sjoberg, Eric, San Diego, CA, UNITED STATES
PATENT ASSIGNEE(S): Cytel Corporation (U.S. corporation)

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 2002119516	A1	20020829
APPLICATION INFO.:	US 2001-7331	A1	20011109 (10)
RELATED APPLN. INFO.:	Division of Ser. No. US 1998-7741, filed on 15 Jan 1998, PENDING		

	NUMBER	DATE
PRIORITY INFORMATION:	US 1997-35710P	19970116 (60)
DOCUMENT TYPE:	Utility	
FILE SEGMENT:	APPLICATION	
LEGAL REPRESENTATIVE:	TOWNSEND AND TOWNSEND AND CREW, LLP, TWO EMBARCADERO CENTER, EIGHTH FLOOR, SAN FRANCISCO, CA, 94111-3834	
NUMBER OF CLAIMS:	58	
EXEMPLARY CLAIM:	1	
NUMBER OF DRAWINGS:	2 Drawing Page(s)	
LINE COUNT:	1150	

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

AB This invention provides methods for practical in vitro sialylation of glycoproteins, including recombinantly produced glycoproteins. The methods are useful for large-scale modification of sialylation patterns.

L10 ANSWER 49 OF 56 USPATFULL on STN
ACCESSION NUMBER: 2002:129751 USPATFULL
TITLE: Practical in vitro sialylation of recombinant glycoproteins
INVENTOR(S): Paulson, James C., Del Mar, CA, United States
Bayer, Robert J., San Diego, CA, United States
Sjoberg, Eric, San Diego, CA, United States
PATENT ASSIGNEE(S): Neose Technologies, Inc., Horsham, PA, United States (U.S. corporation)

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 6399336	B1	20020604
APPLICATION INFO.:	US 1998-7741		19980115 (9)

	NUMBER	DATE
PRIORITY INFORMATION:	US 1997-35710P	19970116 (60)
DOCUMENT TYPE:	Utility	
FILE SEGMENT:	GRANTED	
PRIMARY EXAMINER:	Achutamurthy, Ponnathapu	
ASSISTANT EXAMINER:	Rao, Manjunath N.	
LEGAL REPRESENTATIVE:	Townsend and Townsend and Crew LLP	
NUMBER OF CLAIMS:	87	
EXEMPLARY CLAIM:	1	
NUMBER OF DRAWINGS:	2 Drawing Figure(s); 2 Drawing Page(s)	
LINE COUNT:	1239	

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

AB This invention provides methods for practical in vitro sialylation of glycoproteins, including recombinantly produced glycoproteins. The methods are useful for large-scale modification of sialylation patterns.

L10 ANSWER 50 OF 56 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 2003:30393 BIOSIS
DOCUMENT NUMBER: PREV200300030393
TITLE: Evolution of lipopolysaccharide glycosyltransferase specificity examined by the study of homologous enzymes.
AUTHOR(S): Wakarchuk, Warren W. [Reprint Author]; Bernatchez, Stephane [Reprint Author]; Gilbert, Michel [Reprint Author]; Karwaski, Marie-France [Reprint Author]; Masson, Amara [Reprint Author]; Logan, Susan [Reprint Author]; Dunn, Jessica [Reprint Author]
CORPORATE SOURCE: Institute for Biological Sciences, National Research Council of Canada, 100 Sussex Drive, Ottawa, ON, K1A 0R6, Canada
SOURCE: Glycobiology, (October 2002) Vol. 12, No. 10, pp. 705-706. print.
Meeting Info.: 7th Annual Conference of the Society for Glycobiology. Boston, MA, USA. November 09-12, 2002. Society for Glycobiology.
ISSN: 0959-6658.
DOCUMENT TYPE: Conference; (Meeting)
Conference; (Meeting Poster)
Conference; Abstract; (Meeting Abstract)
LANGUAGE: English
ENTRY DATE: Entered STN: 8 Jan 2003
Last Updated on STN: 11 Feb 2003

L10 ANSWER 51 OF 56 CAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2002:37514 CAPLUS
DOCUMENT NUMBER: 137:16281
TITLE: The genetic bases for the variation in the lipo-oligosaccharide of the mucosal pathogen, **Campylobacter jejuni**. Biosynthesis of sialylated ganglioside mimics in the core oligosaccharide
AUTHOR(S): Gilbert, Michel; Karwaski, Marie-France; Bernatchez, Stephane; Young, N. Martin; Taboada, Eduardo; Michniewicz, Joseph; Cunningham, Anna-Maria; Wakarchuk, Warren W.
CORPORATE SOURCE: Institute for Biological Sciences, National Research Council of Canada, Ottawa, ON, K1A 0R6, Can.
SOURCE: Journal of Biological Chemistry (2002), 277(1), 327-337
CODEN: JBCHA3; ISSN: 0021-9258
PUBLISHER: American Society for Biochemistry and Molecular Biology
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The lipo-oligosaccharide (LOS) biosynthesis loci from 11 **Campylobacter jejuni** strains expressing a total of 8 different ganglioside mimics in their LOS outer cores were compared. Based on the organization of the genes, the 11 corresponding loci could be classified into 3 classes, with one of them being clearly an intermediate evolutionary step between the other two. Comparative genomics and expression of specific glycosyltransferases combined with in vitro activity assays allowed identification of ≥5 distinct mechanisms that allow *C. jejuni* to vary the structure of the LOS outer core as follows: (1) different gene complements; (2) phase variation because of

homopolymeric tracts; (3) gene inactivation by the deletion or insertion of a single base (without phase variation); (4) single mutation leading to the inactivation of a glycosyltransferase; and (5) single or multiple mutations leading to "allelic" glycosyltransferases with different acceptor specificities. The differences in the LOS outer core structures expressed by the 11 *C. jejuni* strains examined can be explained by one or more of these 5 mechanisms.

REFERENCE COUNT: 36 THERE ARE 36 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 52 OF 56 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on STN
ACCESSION NUMBER: 2001:339631 SCISEARCH
THE GENUINE ARTICLE: 423VN
TITLE: Dependence of the bi-functional nature of a **sialyltransferase** from *Neisseria meningitidis* on a single amino acid substitution
AUTHOR: Wakarchuk W W (Reprint); Watson D; St Michael F; Li J J; Wu Y Y; Brisson J R; Young N M; Gilbert M
CORPORATE SOURCE: Natl Res Council Canada, Inst Biol Sci, Immunochem Program, 100 Sussex Dr, Ottawa, ON K1A 0R6, Canada (Reprint); Natl Res Council Canada, Inst Biol Sci, Immunochem Program, Ottawa, ON K1A 0R6, Canada
COUNTRY OF AUTHOR: Canada
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (20 APR 2001) Vol. 276, No. 16, pp. 12785-12790.
Publisher: AMER SOC BIOCHEMISTRY MOLECULAR BIOLOGY INC, 9650 ROCKVILLE PIKE, BETHESDA, MD 20814 USA.
ISSN: 0021-9258.
DOCUMENT TYPE: Article; Journal
LANGUAGE: English
REFERENCE COUNT: 27

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB The L1 immunotype strain 126E of *Neisseria meningitidis* has been shown to have an N-acetyl-neuraminic acid-containing lipooligosaccharide in which an alpha -linked galactose from a pk epitope is substituted at the O6 position (Wakarchuk, W., W., Gilbert, M., Martin, A., Wu, Y., Brisson, J., R., Thibault, P., and Richards, J., C., (1998) Eur. J. Biochem. 254, 626-633). Using a synthetic pk-epitope containing acceptor in glycosyltransferase reactions, we were able to show by NMR analysis of the reaction product that the 126E(L1)-derived **sialyltransferase** can make both **alpha -2,3** and **alpha -2,6** linkages to the terminal galactose. Gene disruption experiments showed that the Ist gene in 126E(L1) was responsible for the in vivo addition of the alpha -2,6-linked N-acetyl-neuraminic acid residue. By site-directed mutagenesis it was possible to change the MC58(L3)-derived enzyme into a bifunctional enzyme with a single amino acid change at position 168, where a glycine was changed to an isoleucine. We performed a gene replacement experiment where the 126E(L1) **alpha -2,3/6-sialyltransferase** was replaced by allelic exchange with the monofunctional MC58(L3) **alpha -2,3-sialyltransferase** and with the mutant MC58(L3) allele G168I. We observed that the level of LOS sialylation with the G168I allele was very similar to that of the wild type 126E(L1), indicating that residue 168 is the critical residue for the **alpha -2,6-sialyltransferase** activity in vitro as well as in vivo.

L10 ANSWER 53 OF 56 CAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 2000:553711 CAPLUS
DOCUMENT NUMBER: 133:161277
TITLE: Campylobacter glycosyltransferases for biosynthesis of gangliosides and ganglioside mimics
INVENTOR(S): Gilbert, Michel; Wakarchuk, Warren W.

PATENT ASSIGNEE(S): National Research Council of Canada, Can.
SOURCE: PCT Int. Appl., 120 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 3
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2000046379	A1	20000810	WO 2000-CA86	20000201
W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, ZA, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
US 6503744	B1	20030107	US 2000-495406	20000131
CA 2360205	AA	20000810	CA 2000-2360205	20000201
EP 1147200	A1	20011024	EP 2000-901455	20000201
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, LT, LV, FI, RO				
JP 2002535992	T2	20021029	JP 2000-597438	20000201
AU 772569	B2	20040429	AU 2000-22743	20000201
PRIORITY APPLN. INFO.:				
			US 1999-118213P	P 19990201
			US 2000-495406	A 20000131
			WO 2000-CA86	W 20000201

AB This invention provides prokaryotic glycosyltransferases, including a bifunctional sialyltransferase that has both an α 1,3- and an α 2,8- activity. A β 1,4-GalNAc transferase and a β 1,3-galactosyltransferase are also provided by the invention, as are other glycosyltransferases and enzymes involved in synthesis of lipooligosaccharide (LOS). The glycosyltransferases can be obtained from, for example, *Campylobacter* species, including *C. jejuni*. In addnl. embodiments, the invention provides nucleic acids that encode the glycosyltransferases, as well as expression vectors and host cells for expressing the glycosyltransferases.

REFERENCE COUNT: 11 THERE ARE 11 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 54 OF 56 CAPLUS COPYRIGHT 2005 ACS on STN DUPLICATE 6
ACCESSION NUMBER: 2000:129311 CAPLUS
DOCUMENT NUMBER: 132:304980
TITLE: Biosynthesis of ganglioside mimics in *Campylobacter jejuni* OH4384. Identification of the glycosyltransferase genes, enzymatic synthesis of model compounds, and characterization of nanomole amounts by 600-MHz H and C NMR analysis
AUTHOR(S): Gilbert, Michel; Brisson, Jean-Robert; Karwaski, Marie-France; Michniewicz, Joseph; Cunningham, Anna-Maria; Wu, Yuyang; Young, N. Martin; Wakarchuk, Warren W.
CORPORATE SOURCE: Institute for Biological Sciences, National Research Council of Canada, Ottawa, ON, K1A 0R6, Can.
SOURCE: Journal of Biological Chemistry (2000), 275(6), 3896-3906
CODEN: JBCHA3; ISSN: 0021-9258
PUBLISHER: American Society for Biochemistry and Molecular Biology
DOCUMENT TYPE: Journal

LANGUAGE: English

AB We have applied two strategies for the cloning of four genes responsible for the biosynthesis of the GT1a ganglioside mimic in the lipooligosaccharide (LOS) of a bacterial pathogen, **Campylobacter jejuni** OH4384, which has been associated with Guillain-Barre syndrome. We first cloned a gene encoding an **.alpha.-2,3-sialyltransferase** (cst-I) using an activity screening strategy. We then used nucleotide sequence information from the recently completed sequence from *C. jejuni* NCTC 11168 to amplify a region involved in LOS biosynthesis from *C. jejuni* OH4384. The LOS biosynthesis locus from *C. jejuni* OH4384 is 11.47 kilobase pairs and encodes 13 partial or complete open reading frames, while the corresponding locus in *C. jejuni* NCTC 11168 spans 13.49 kilobase pairs and contains 15 open reading frames, indicating a different organization between these two strains. Potential glycosyltransferase genes were cloned individually, expressed in *Escherichia coli*, and assayed using synthetic fluorescent oligosaccharides as acceptors. We identified genes encoding a β -1,4-N-acetylgalactosaminyltransferase (cgtA), a β -1,3-galactosyltransferase (cgtB), and a bifunctional **sialyltransferase** (cst-II), which transfers sialic acid to O-3 of galactose and to O-8 of a sialic acid that is linked **.alpha.-2,3-** to a galactose. The linkage specificity of each identified glycosyltransferase was confirmed by NMR anal. at 600 MHz on nanomole amts. of model compds. synthesized in vitro. Using a gradient inverse broadband nano-NMR probe, sequence information could be obtained by detection of 3J(C,H) correlations across the glycosidic bond. The role of cgtA and cst-II in the synthesis of the GT1a mimic in *C. jejuni* OH4384 were confirmed by comparing their sequence and activity with corresponding homologs in two related *C. jejuni* strains that express shorter ganglioside mimics in their LOS.

REFERENCE COUNT: 31 THERE ARE 31 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 55 OF 56 CAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1999:626342 CAPLUS

DOCUMENT NUMBER: 131:253359

TITLE: **Campylobacter jejuni** gene cst-I

lipopolysaccharide **.alpha.-2,**

3 sialyltransferase, its DNA and

amino acid sequences, recombinant production, and its acceptor specificity

INVENTOR(S): Gilbert, Michel; Wakarchuk, Warren W.

PATENT ASSIGNEE(S): National Research Council of Canada, Can.

SOURCE: PCT Int. Appl., 47 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9949051	A1	19990930	WO 1999-CA238	19990322
W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
US 6689604	B1	20040210	US 1999-272960	19990318
CA 2323753	AA	19990930	CA 1999-2323753	19990322

AU 9928230	A1	19991018	AU 1999-28230	19990322
AU 745040	B2	20020307		
EP 1082440	A1	20010314	EP 1999-908717	19990322
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
JP 2002507424	T2	20020312	JP 2000-538012	19990322
US 2003049270	A1	20030313	US 2002-58636	20020129
US 6709834	B2	20040323		
US 2004152165	A1	20040805	US 2004-799016	20040311
PRIORITY APPLN. INFO.:				
			US 1998-78891P	P 19980320
			US 1999-272960	A 19990318
			WO 1999-CA238	W 19990322
			US 2002-58636	A3 20020129

AB The invention provides DNA mols. that encode gene cst-I lipopolysaccharide .alpha.-2,3 sialyltransferase of *Campylobacter jejuni*. The DNA sequence of *C. jejuni* gene cst-I, as well as the corresponding amino acid sequence of lipopolysaccharide .alpha.-2,3 sialyltransferase are claimed. The invention also provides methods for the recombinant production of lipopolysaccharide .alpha.-2,3 sialyltransferase in prokaryotic and eukaryotic cells. The invention further provides the specificity of the *C. jejuni* lipopolysaccharide .alpha.-2,3 sialyltransferase. The *C. jejuni* lipopolysaccharide .alpha.-2,3 sialyltransferase uses terminal galactose acceptors that are β -(1 \rightarrow 4) linked to either glucose or N-acetylglucosamine. The enzyme also uses terminal galactose acceptors that are β -(1 \rightarrow 3) linked to N-acetylglucosamine or N-acetylgalactosamine. The enzyme uses cytidine monophosphate-N-acetylneuraminic acid (CMP-Neu5Ac) as the donor. The broad acceptor specificity of lipopolysaccharide .alpha.-2,3 sialyltransferase encoded by cst-I demonstrates its utility and makes it an attractive tool for chemo-enzymic synthesis of sialylated oligosaccharides.

REFERENCE COUNT: 4 THERE ARE 4 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 56 OF 56	MEDLINE on STN	DUPPLICATE 7
ACCESSION NUMBER:	1999449955 MEDLINE	
DOCUMENT NUMBER:	PubMed ID: 10520252	
TITLE:	Synthesis of a disialylated hexasaccharide of type VIII group B <i>Streptococcus</i> capsular polysaccharide.	
AUTHOR:	Eichler E; Jennings H J; Gilbert M; Whitfield D M	
CORPORATE SOURCE:	National Research Council, Ottawa, Ontario, Canada.	
SOURCE:	Carbohydrate research, (1999 Jun 30) 319 (1-4) 1-16. Journal code: 0043535. ISSN: 0008-6215.	
PUB. COUNTRY:	Netherlands	
DOCUMENT TYPE:	Journal; Article; (JOURNAL ARTICLE)	
LANGUAGE:	English	
FILE SEGMENT:	Priority Journals	
ENTRY MONTH:	199912	
ENTRY DATE:	Entered STN: 20000113 Last Updated on STN: 20000113 Entered Medline: 19991217	

AB As part of our program to design, develop and prepare protective vaccines against the bacterial pathogens Group B *Streptococcus*, we report the synthesis of a disialylated hexasaccharide. This hexasaccharide represents a portion of the serotype-specific capsular polysaccharide of Type VIII that has the tetrasaccharide repeat unit [β -L-Rhap-(1 \rightarrow 4)- β -D-Glcp-(1 \rightarrow 4)-[α -Neu5Ac-(2 \rightarrow 3)]- β -D-Galp-(1 \rightarrow 4)]n. A tetrasaccharide corresponding to this repeat unit has been synthesized by us [E. Eichler, H.J. Jennings, D.M. Whitfield, J. Carbohydr. Chemical, 16 (1997) 385-411]. Since the protective epitopes are believed to involve

several repeat units, methods to extend this tetrasaccharide were examined. This objective requires a glycosylation of the unreactive OH-4 of the beta-L-Rhap, which was accomplished by coupling a D-Galp glycosyl trichloroacetimidate donor with a beta-L-Rhap-(1-->4)-D-Glcp acceptor. Subsequent coupling of this trisaccharide as a donor to an alpha-Neu5Ac-(2-->3)-D-Galp disaccharide acceptor gave a pentasaccharide. The pentasaccharide was deprotected and enzymatically sialylated using an **alpha-(2-->3)-sialyltransferase** from **Campylobacter jejuni** to give the title hexasaccharide
alpha-Neu5Ac-(2-->3)-beta-D-Galp-(1-->4)-beta-L-Rhap-(1-->4)-beta-D-Glcp-(1-->4)-[alpha-Neu5Ac-(2-->3)]-beta-D-Galp-(1-->0)-(CH₂)₃N₃.

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L4 ANSWER 22 OF 32 USPATFULL on STN
 ACCESSION NUMBER: 2000:98214 USPATFULL
 TITLE: Recombinant α -2,3-sialyltransferases
 and their uses
 INVENTOR(S): Gilbert, Michel, Hull, Canada
 Wakarchuk, Warren W., Gloucester, Canada
 Young, Martin N., Gloucester, Canada
 Jennings, Michael P., Carina, Australia
 PATENT ASSIGNEE(S): National Research Council of Canada, Ottawa, Canada
 (non-U.S. corporation)
 The Chancellor, Masters and Scholars of the University
 of Oxford, Oxford, United Kingdom (non-U.S.
 corporation)

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 6096529		20000801
APPLICATION INFO.:	US 1997-872485		19970607 (8)
DOCUMENT TYPE:		Utility	
FILE SEGMENT:		Granted	
PRIMARY EXAMINER:		Prouty, Rebecca E.	
LEGAL REPRESENTATIVE:		Townsend and Townsend and Crew LLP	
NUMBER OF CLAIMS:	30		
EXEMPLARY CLAIM:	1		
NUMBER OF DRAWINGS:	3	Drawing Figure(s); 2 Drawing Page(s)	
LINE COUNT:	1864		
CAS INDEXING IS AVAILABLE FOR THIS PATENT.			
AB	The structure and specificity of the recombinant α -2,3-sialyltransferases from <i>Neisseria</i> spp, are disclosed. Their use in the production of desired carbohydrate structures is also provided.		

L4 ANSWER 23 OF 32 CAPLUS COPYRIGHT 2005 ACS on STN DUPLICATE 2
 ACCESSION NUMBER: 1999:354320 CAPLUS
 DOCUMENT NUMBER: 131:31097
 TITLE: Method of the production of saccharides containing sialic acid
 INVENTOR(S): Yamamoto, Takeshi; Nakashizuka, Motoko; Terada, Ichiro; Kodama, Hisashi
 PATENT ASSIGNEE(S): Japan Tobacco Inc., Japan
 SOURCE: U.S., 15 pp.
 CODEN: USXXAM
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 2
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 5908766	A	19990601	US 1996-741663	19961031
JP 08173182	A2	19960709	JP 1994-327152	19941228
JP 3124199	B2	20010115		

PRIORITY APPLN. INFO.: JP 1994-327152 A 19941228
 OTHER SOURCE(S): CASREACT 131:31097
 AB Described is a method of production of saccharides containing sialic acid, wherein

β -galactoside- α -2,6-sialyltransferase is used for linking sialic acid to the 6-position of a galactose residue in a sugar chain of a glycoconjugate or the 6-position of a galactose residue in a free sugar chain, or to the 6-position of a monosaccharide having an OH group at C6 and being capable of forming an oligosaccharide or a glycoconjugate.

REFERENCE COUNT: 13 THERE ARE 13 CITED REFERENCES AVAILABLE FOR THIS

RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 24 OF 32 CAPLUS COPYRIGHT 2005 ACS on STN
 ACCESSION NUMBER: 1998:585449 CAPLUS
 DOCUMENT NUMBER: 129:272316
 TITLE: Novel signal peptide of a marine bacterial
 β -galactoside . alpha.2,
 6-sialyltransferase gene from
Photobacterium damsela JT0160
 INVENTOR(S): Yamamoto, Takeshi; Nakashizu, Motoko; Terada, Ichiro
 PATENT ASSIGNEE(S): Japan Tobacco, Inc., Japan
 SOURCE: Jpn. Kokai Tokkyo Koho, 25 pp.
 CODEN: JKXXAF
 DOCUMENT TYPE: Patent
 LANGUAGE: Japanese
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 10234374	A2	19980908	JP 1997-45103	19970228
PRIORITY APPLN. INFO.:			JP 1997-45103	19970228

AB **Sialyltransferase** 0160, a bacterial **sialyltransferase**
 which catalyzes the incorporation of NeuAc from CMP-NeuAc into the
 galactose residue of the carbohydrate chain at position 6, is produced by
Photobacterium damsela JT0160. The gene coding for
sialyltransferase 0160 (bst) was cloned, sequenced, and its signal
 sequence identified.

L4 ANSWER 25 OF 32 CAPLUS COPYRIGHT 2005 ACS on STN
 ACCESSION NUMBER: 1998:585448 CAPLUS
 DOCUMENT NUMBER: 129:213508
 TITLE: Cloning and expression of a marine bacterial
 β -galactoside . alpha.2,
 6-sialyltransferase gene from
Photobacterium damsela JT0160
 INVENTOR(S): Yamamoto, Takeshi; Nakashizu, Motoko; Terada, Ichiro
 PATENT ASSIGNEE(S): Japan Tobacco, Inc., Japan
 SOURCE: Jpn. Kokai Tokkyo Koho, 26 pp.
 CODEN: JKXXAF
 DOCUMENT TYPE: Patent
 LANGUAGE: Japanese
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 10234373	A2	19980908	JP 1997-45087	19970228
JP 3140976	B2	20010305		
CA 2252493	AA	19980903	CA 1998-2252493	19980302
WO 9838315	A1	19980903	WO 1998-JP850	19980302
W: CA, US				
RW: DE, FR, GB				
EP 915163	A1	19990512	EP 1998-905721	19980302
R: DE, FR, GB				
US 6255094	B1	20010703	US 1999-171878	19990105
PRIORITY APPLN. INFO.:			JP 1997-45087	A 19970228
			WO 1998-JP850	W 19980302

AB **Sialyltransferase** 0160, a bacterial **sialyltransferase**
 which catalyzes the incorporation of NeuAc from CMP-NeuAc into the
 galactose residue of the carbohydrate chain at position 6, is produced by
Photobacterium damsela JT0160. The gene coding for
sialyltransferase 0160 (bst) was cloned, sequenced, and expressed

in *Escherichia coli*. Claimed is a method for the recombinant preparation of the enzyme.

L4 ANSWER 26 OF 32 CAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 1998:585440 CAPLUS
DOCUMENT NUMBER: 129:256942
TITLE: Cloning and expression of gene for
β-galactoside-. alpha.2,
6-sialyltransferase from marine
bacteria *Photobacterium damsela*
INVENTOR(S): Yamamoto, Takeshi; Nakashizu, Motoko; Terada, Ichiro
PATENT ASSIGNEE(S): Japan Tobacco, Inc., Japan
SOURCE: Jpn. Kokai Tokkyo Koho, 26 pp.
CODEN: JKXXAF
DOCUMENT TYPE: Patent
LANGUAGE: Japanese
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 10234364	A2	19980908	JP 1997-45098	19970228
CA 2252561	AA	19980903	CA 1998-2252561	19980302
WO 9838289	A1	19980903	WO 1998-JP851	19980302
W: CA, US				
RW: DE, FR, GB				
EP 915153	A1	19990512	EP 1998-905722	19980302
R: DE, FR, GB				
PRIORITY APPLN. INFO.:			JP 1997-45098	A 19970228
			WO 1998-JP851	W 19980302

AB The gene for a novel 675-amino-acid β-galactoside-. alpha.2, 6-sialyltransferase is isolated from *Photobacterium damsela* strain JT0160 and expressed in *Escherichia coli*. Claimed are the signal sequence encoding the signal peptide (amino acids 1-15) of the enzyme, the soluble form of the enzyme (amino acids 16-498), and a recombinant method for the preparation of the enzyme.

L4 ANSWER 27 OF 32 USPATFULL on STN
ACCESSION NUMBER: 1998:131590 USPATFULL
TITLE: β-galactoside-. alpha.-2,
6-sialyltransferase, and a process
for producing from *Photobacterium*
INVENTOR(S): Yamamoto, Takeshi, Kanagawa, Japan
Nakashizuka, Motoko, Kanagawa, Japan
Terada, Ichiro, Osaka, Japan
Kodama, Hisashi, Tokyo, Japan
PATENT ASSIGNEE(S): Japan Tobacco Inc., Tokyo, Japan (non-U.S. corporation)

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 5827714		19981027
APPLICATION INFO.:	US 1996-739015		19961028 (8)
DOCUMENT TYPE:	Utility		
FILE SEGMENT:	Granted		
PRIMARY EXAMINER:	Weber, Jon P.		
LEGAL REPRESENTATIVE:	Birch, Stewart, Kolasch & Birch, LLP		
NUMBER OF CLAIMS:	12		
EXEMPLARY CLAIM:	1		
NUMBER OF DRAWINGS:	9 Drawing Figure(s); 9 Drawing Page(s)		
LINE COUNT:	786		
CAS INDEXING IS AVAILABLE FOR THIS PATENT.			

AB A β-galactoside-. alpha.-2, 6-

sialyltransferase derived from a microorganism belonging to the genus *Photobacterium* has been disclosed, having the following physicochemical properties:

- (1) action and specificity: transferring sialic acid from cytidine monophosphate-sialic acid to the 6-position of a galactose residue in a sugar chain of a glycoconjugate or in a free sugar chain, or to the 6-position of a monosaccharide having a hydroxyl group on carbon at the 6-position and being capable of forming a glycoconjugate.
- (2) optimum pH: 5 to 6;
- (3) optimum temperature: 30° C.; and
- (4) molecular weight: 64,000±5,000 (determined by gel filtration).

L4 ANSWER 28 OF 32 CAPLUS COPYRIGHT 2005 ACS on STN DUPLICATE 3
ACCESSION NUMBER: 1998:174532 CAPLUS
DOCUMENT NUMBER: 128:256425
TITLE: Mass production of bacterial α .
 α .2,6-sialyltransferase and enzymic syntheses of sialyloligosaccharides
AUTHOR(S): Yamamoto, Takeshi; Nagae, Hideki; Kajihara, Yasuhiro;
Terada, Ichiro
CORPORATE SOURCE: Sea Water Science Research Laboratory, Japan Tobacco
Inc., Kanagawa, 256, Japan
SOURCE: Bioscience, Biotechnology, and Biochemistry (1998),
62(2), 210-214
CODEN: BBBIEJ; ISSN: 0916-8451
PUBLISHER: Japan Society for Bioscience, Biotechnology, and
Agrochemistry
DOCUMENT TYPE: Journal
LANGUAGE: English
AB To supply α .2,6-sialyltransferase for the large-scale synthesis of sialoside, the authors investigated culture conditions for the production of **sialyltransferase** 0160. The addition of galactose and beef extract and control of the pH of the culture medium were effective on the production of **sialyltransferase** 0160. The maximal enzyme productivity reached 550 units/L. Using a crude extract of **Photobacterium damsela** JT0160 cells as an enzyme source, enzymic syntheses were performed with mono- and di-saccharides as the sialyl acceptors. It was clarified that a crude extract of **P. damsela** JT0160 cells can be used as an synthetic catalyst for the enzymic synthesis of sialyloligosaccharides. Furthermore, the enzyme assay showed that **sialyltransferase** 0160 could transfer NeuAc to not only N-linked but also O-linked carbohydrate chains. These results indicated that an abundant supply of **sialyltransferase** 0160 and its broad specificity make possible the synthesis of sialoside on a large scale.
REFERENCE COUNT: 21 THERE ARE 21 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 29 OF 32 CAPLUS COPYRIGHT 2005 ACS on STN DUPLICATE 4
ACCESSION NUMBER: 1998:145210 CAPLUS
DOCUMENT NUMBER: 128:279370
TITLE: Cloning and expression of a marine bacterial β -galactoside α .2,6-sialyltransferase gene from **Photobacterium damsela** JT0160
AUTHOR(S): Yamamoto, Takeshi; Nakashizuka, Motoko; Terada, Ichiro
CORPORATE SOURCE: Sea Water Science Research Laboratory, Japan Tobacco
Inc., Kanagawa, 256, Japan
SOURCE: Journal of Biochemistry (Tokyo) (1998), 123(1), 94-100

CODEN: JOBIAO; ISSN: 0021-924X
PUBLISHER: Japanese Biochemical Society
DOCUMENT TYPE: Journal
LANGUAGE: English
AB **Sialyltransferase 0160**, a bacterial **sialyltransferase** which catalyzes the incorporation of NeuAc from CMP-NeuAc into the galactose residue of the carbohydrate chain at position 6, is produced by **Photobacterium damsela** JT0160. The gene coding for **sialyltransferase 0160** (bst) was cloned, sequenced, and expressed in **Escherichia coli**. The **sialyltransferase 0160** gene contains an open reading frame of 2,028 base pairs encoding a protein of 675 amino acid residues. The deduced amino acid sequence of **sialyltransferase 0160** did not contain the sialylmotif and had no significant similarity to mammalian **sialyltransferases**. Crude exts. of cultured **E. coli** MV1184 cells carrying an expression plasmid for the **sialyltransferase 0160** gene showed **sialyltransferase** activity, which was identified as β -galactoside . **alpha**.
2,6-sialyltransferase activity by enzymic reaction product anal. In addition, when mutant genes, lacking 3'-coding regions for COOH-terminal portions of the protein, which are thought to form α -helix structures, were expressed in **E. coli** MV1184, soluble-form enzymes were obtained. This implies that the COOH-terminal portion of **sialyltransferase 0160** is required for membrane binding.

REFERENCE COUNT: 39 THERE ARE 39 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 30 OF 32 CAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 1996:501582 CAPLUS
DOCUMENT NUMBER: 125:162103
TITLE: β -Galactoside-. **alpha**.2,
6-sialyltransferase of
Photobacterium damsela, its preparation, and use for manufacturing complex sugars
INVENTOR(S): Yamamoto, Takeshi; Nakashizu, Motoko; Terada, Ichiro;
Kodama, Hisashi
PATENT ASSIGNEE(S): Nippon Tobacco Sangyo, Japan
SOURCE: Jpn. Kokai Tokkyo Koho, 10 pp.
CODEN: JKXXAF
DOCUMENT TYPE: Patent
LANGUAGE: Japanese
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 08154673	A2	19960618	JP 1994-304090	19941207
JP 3062409	B2	20000710		
<u>US 5827714</u>	A	19981027	US 1996-739015	19961028

PRIORITY APPLN. INFO.: JP 1994-304090 A 19941207
AB A method for the preparation of β -galactoside-. **alpha**.2,
6-sialyltransferase from the culture of **Photobacterium damsela** strains JT0160, ATCC 33539, and ATCC 35083 is disclosed. The enzyme prepared from **P. damsela** strain exhibits a pH optimum 5, temperature optimum 30°, pI 4.6, and mol. weight 61,000 by SDS-PAGE or 64,000 by gel filtration. Manufacture of sialylmethyl- β -D-N-acetyllactosamine from methyl- β -D-N-acetyllactosamine in the presence of the enzyme was demonstrated.

L4 ANSWER 31 OF 32 CAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 1996:674396 CAPLUS
DOCUMENT NUMBER: 125:321393
TITLE: A Novel .**alpha**.-2,6-
Sialyltransferase: Transfer of Sialic Acid to

AUTHOR(S) : Fucosyl and Sialyl Trisaccharides
Kajihara, Yasuhiro; Yamamoto, Takeshi; Nagae, Hideki;
Nakashizuka, Motoko; Sakakibara, Tohru; Terada, Ichiro
CORPORATE SOURCE: Department of System Function, Yokohama City
University, Yokohama, 236, Japan
SOURCE: Journal of Organic Chemistry (1996), 61(24), 8632-8635
CODEN: JOCEAH; ISSN: 0022-3263
PUBLISHER: American Chemical Society
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The substrate specificity and enzymic sialylation ability of the bacterium .
.alpha.-2,6-sialyltransferase were
examined. The enzyme assay displayed a remarkable ability to catalyze sialyl
transfer to type-II oligosaccharides possessing fucosides or sialosides at
the 2 or 3 position of the terminal galactoside. Enzymic syntheses were
performed to confirm the structure of unusual assay products found when
using Neu5Ac β 2,3Gal β 1,4Glc and Fuc α 1,2Gal β 1,4Glc as
the sialyl acceptors. Both sialylation reactions (10 μ mol scales) were
run using 83 munits of enzyme, were complete in 2 h, and afforded the
sialoside analogs Neu5Ac .
.alpha.2,6(Fuc
 α 1,2) Gal β 1,4Glc (88%) and Neu5Ac .
.alpha.2,
6(Neu5Ac β 2,3) Gal β 1,4Glc (92%).

L4 ANSWER 32 OF 32 CAPLUS COPYRIGHT 2005 ACS on STN DUPLICATE 5
ACCESSION NUMBER: 1996:515656 CAPLUS
DOCUMENT NUMBER: 125:161823
TITLE: Purification and characterization of a marine
bacterial β -galactoside .
.alpha.
2,6-sialyltransferase from
Photobacterium damsela JT0160
AUTHOR(S) : Yamamoto, Takeshi; Nakashizuka, Motoko; Kodama,
Hisashi; Kajihara, Yasuhiro; Terada, Ichiro
CORPORATE SOURCE: Japan Tobacco Inc., Sea Water Sci. Res. Lab.,
Kanagawa, 256, Japan
SOURCE: Journal of Biochemistry (Tokyo) (1996), 120(1),
104-110
CODEN: JOBIAO; ISSN: 0021-924X
PUBLISHER: Japanese Biochemical Society
DOCUMENT TYPE: Journal
LANGUAGE: English
AB A bacterial sialyltransferase, designated
sialyltransferase 0160, was purified from a marine bacterium that
had been isolated from seawater from Sagami Bay, Kanagawa. This strain
was identified as *P. damsela*, and designated *P. damsela* JT0160.
Sialyltransferase 0160 was purified 688-fold to homogeneity from
the crude extract of the cells with a yield of 19% using a combination of
anion-exchange chromatog., hydroxylapatite chromatog., gel filtration
chromatog., and affinity chromatog. The purified enzyme migrated as a
single band (61 kDa) in SDS-PAGE. This sialyltransferase was
found to be a β -galactoside .
.alpha.2,6-
sialyltransferase (EC 2.4.99.1), which catalyzes the incorporation
of NeuAc from CMP-NeuAc into the galactose residue of the carbohydrate
chain at position 6, on the basis of an anal. of the enzymic reaction
products with HPLC, 1 H-NMR, 13 C-NMR spectroscopy, and fast atom
bombardment mass spectroscopy.

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